

SUPPLEMENTARY FEEDING ON THE NUTRIENT BALANCE OF LACTATING DAIRY COW AT CONTRASTING TEMPERATURE REGIMES: ASSESSMENT USING CORNELL NET CARBOHYDRATE AND PROTEIN SYSTEM (CNCPS) MODEL

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ABSTRACT

Dairy cows often do not receive adequate nutrient supply during their lactation period. This condition can even be worse if the environmental temperature is not in comfortable range which may occur especially in tropical regions. The present research was aimed to simulate the effect of supplementary feeding on nutrient balance of lactating dairy cow at contrasting temperature regimes using Cornell Net Carbohydrate and Protein System (CNCPS) model. Treatments consisted of feeds (R1: *Pennisetum purpureum*, R2: *P. purpureum* + concentrate (60:40), R3: *P. purpureum* + *Gliricidia sepium* + *Leucaena leucocephala* (60:20:20), R4: *P. purpureum* + concentrate + *G. sepium* + *L. leucocephala* (60:20:10:10)) and environmental temperatures (T1: 20 °C, T2: 30 °C). The dairy cow inputs in CNCPS were Holstein breed, body weight of 500 kg, feed intake of 15 kg (dry matter basis) per day and produced milk 15 kg/day. Based on the CNCPS model, there were negative balances of metabolisable energy (ME) and metabolisable protein (MP) if a lactating dairy cow fed only by *P. purpureum*. The ME balance was worse at higher temperature, while the MP balance was remain unchanged. Addition of concentrate mixture (R2) fulfilled the ME and MP requirements as well as other nutrients. Addition of leguminous tree leaves (R3 and R4) improved the nutritional status of the lactating cow model compared to R1, but did not better than R2. It was concluded that supplementary feeding is necessary for improving the nutrient balance of lactating dairy cow, especially when the cow is maintained under uncomfortable environmental temperature.

Keywords: *cncps, Supplementation, Temperature, Nutrient Balance*

INTRODUCTION

Dairy cows often do not receive adequate nutrient supply during their lactation period. Metabolizable energy (ME) intake is considered as the first limiting factor for milk production of cows fed forage diet, and supply of protein to the duodenum has also been proposed to limit milk production especially for high producing cows (Kolver and Muller, 1998). This condition can even be worse if the environmental temperature is not in comfortable range which may occur, especially in tropical regions. Adverse effects of environmental stress on production of lactating

dairy cows have been well characterized, such as reduced feed intake, growth or milk production, efficiency and reproduction (Thatcher, 1974; Hahn, 1999). Therefore, inadequate nutrient supply may be more severe under dairy production system in the tropics.

To avoid the problem of inadequate nutrient supply, supplementary feeding is necessary. The main objective of supplementation of lactating dairy cows is to increase total dry matter intake, energy intake and nutrient supply relative to that achieved with forage-only diets (Stockdale, 2000; Bargo *et al.*, 2003). For the production system, a primary goal of

supplementation is to optimize profit per cow and per unit of land (Fales *et al.*, 1995). The objectives of supplementation include: (1) increase milk production per cow, (2) increase stocking rate and milk production per unit of land, (3) improve the use of forage with higher stocking rate, (4) maintain or improve body condition score to improve reproduction during forage shortage, (5) increase length of lactation during periods of forage shortage, and (6) increase milk protein content by energy supplementation (Kellaway and Porta, 1993). In relation to these, evaluation of supplementary feeding practices whether they are able to reach the objectives mentioned is also important and can not be ignored.

Cornell Net Carbohydrate and Protein System (CNCPS) is a model designed to evaluate diets and animal performance for all classes of cattle in a unique production situations, using science-based principles of rumen function, microbial growth, feed digestion and passage and physiological state (Fox *et al.*, 2004).

different temperature regimes using the CNCPS model. Other related variables were also simulated, i.e. rumen values and manure excretion. Supplementation was done either in the form of concentrate or leguminous tree leaves (*Gliricidia sepium* and *Leucaena leucocephala*) or mixture of concentrate and the legumes.

MATERIALS AND METHODS

Location and Animal Experiments

Prior to model running, location and animal inputs were defined according to common production systems in Indonesia for dairy cows. The dairy cow inputs in CNCPS were Holstein breed, body weight of 500 kg, feed intake of 15 kg (dry matter basis) per day and produced milk 15 kg/day. The summary of location and animal inputs are presented in Table 1.

Table 1. Location and Animal Inputs for Running The Simulated Treatments

| Variable | Input |
|--------------------------------|--------------------------------------|
| <i>Location</i> | |
| Location type | Small free-stall |
| Temperature (°C) | According to treatment (20 or 30 °C) |
| Minimum night temperature (°C) | According to treatment (15 or 22 °C) |
| Relative humidity (%) | 80 |
| Hours standing | 12 |
| <i>Animal</i> | |
| Animal type | Lactating dairy cow |
| Breed | Holstein |
| Body weight (kg) | 500 |
| Age (months) | 60 |
| Lactation number | 3 |
| Age at first calving (months) | 24 |
| Dry matter intake (kg/day) | 15 |
| Milk production (kg/day) | 15 |
| Milk fat (%) | 3.7 |
| Milk protein (%) | 3.3 |
| Milk lactose (%) | 4.8 |

Therefore, by accounting for farm-specific animal, feed and environmental characteristics, more accurate prediction of dietary nutrient requirements for maintenance, growth and milk production of cattle and nutrient excretion in diverse production situations is possible (Tylutki *et al.*, 2008). The objective of this study was to simulate the effect of supplementary feeding on nutrient balance of lactating dairy cow at

Treatments

Treatments consisted of two factors, i.e. feeds (4 treatments) and environmental temperatures (2 treatments). Therefore, by factorial design there were 8 simulated treatments in total. The temperature treatments consisted of 20°C (comfortable temperature) and 30°C (under stress). The feed treatments consisted of:

R1: Napier grass or *Pennisetum purpureum* (control)

R2: *P. purpureum* + concentrate (60:40, dry matter basis)

R3: *P. purpureum* + *Gliricidia sepium* + *Leucaena leucocephala* (60:20:20)

R4: *P. purpureum* + concentrate + *G. sepium* + *L. leucocephala* (60:20:10:10)

Model Running

The above-mentioned location and animal inputs and treatments were implemented in the model software, and the model was run. The output variables were rumen values, nutrient balance and excretion of the nutrients.

RESULTS AND DISCUSSION

Rumen Values

Rumen values of the simulated treatments are presented in Table 3. The CNCPS divides the ruminal microbial ecosystem into two microbial groups, i.e. microbes that ferment fiber carbohydrate (FC) and those that ferment non fiber carbohydrate (NFC). This segregation reflects differences in N utilization and growth efficiency as well as an almost exclusive partition of energy source utilization. The FC bacteria

Concentrate mixture comprised of (in dry matter basis): cassava residue (33.3%), corn (16.7%), rice bran (16.7%), soybean meal (16.7%), fish meal (5%), corn oil (5%), molasses (3.2%), urea (1.7%) and dicalcium phosphate (1.7%). All the chemical constituents of the feeds used were originated from the CNCPS library. Chemical constituents of feeds used in the simulated treatments are presented in Table 2.

Table 2. Chemical Composition of Feeds Used in The Simulated Treatments

| Component | R1 | Concentrate | <i>G. sepium</i> | <i>L.leucocephala</i> | R2 | R3 | R4 |
|-----------------|-------|-------------|------------------|-----------------------|-------|-------|-------|
| CP (%DM) | 6.60 | 21.04 | 24.40 | 11.70 | 12.37 | 11.18 | 11.78 |
| RUP (%CP) | 65.97 | 30.59 | 38.73 | 58.31 | 47.37 | 52.44 | 49.70 |
| RDP (%CP) | 34.03 | 69.41 | 61.27 | 41.69 | 52.63 | 47.56 | 50.30 |
| Sol P (%CP) | 46.00 | 45.15 | 31.60 | 25.00 | 45.42 | 35.32 | 40.63 |
| ME (Mcal/kg) | 1.40 | 3.26 | 2.27 | 1.43 | 2.11 | 1.58 | 1.84 |
| NEI (Mcal/kg) | 0.90 | 2.10 | 1.46 | 0.92 | 1.36 | 1.02 | 1.19 |
| NDF (%DM) | 74.00 | 17.38 | 37.30 | 64.30 | 51.35 | 64.72 | 58.04 |
| peNDF (%DM) | 44.40 | 9.40 | 35.44 | 57.87 | 30.40 | 45.30 | 37.85 |
| Lignin (%NDF) | 9.60 | 11.23 | 24.40 | 21.40 | 10.25 | 14.92 | 12.59 |
| NFC (%DM) | 7.85 | 49.61 | 26.50 | 18.50 | 24.55 | 13.71 | 19.13 |
| Sugar (%DM) | 7.22 | 15.21 | 21.00 | 10.00 | 10.42 | 10.53 | 10.47 |
| Starch (%DM) | 0.63 | 19.71 | 1.50 | 7.00 | 8.26 | 2.08 | 5.17 |
| Sol fiber (%DM) | 0.00 | 14.55 | 4.00 | 1.50 | 5.82 | 1.10 | 3.46 |
| EE (%DM) | 2.30 | 9.49 | 3.20 | 0.70 | 5.18 | 2.16 | 3.67 |
| LCFA (%DM) | 1.39 | 8.31 | 1.98 | 0.43 | 4.16 | 1.32 | 2.74 |
| Ash (%DM) | 9.40 | 6.74 | 8.60 | 4.80 | 8.33 | 8.32 | 8.33 |
| Ca (%DM) | 0.51 | 0.72 | 0.00 | 2.28 | 0.60 | 0.76 | 0.68 |
| P (%DM) | 0.28 | 1.00 | 0.00 | 0.20 | 0.57 | 0.21 | 0.39 |

R1, *P. purpureum*; R2, *P. purpureum* + concentrate (60:40); R3, *P. purpureum* + *G. sepium* + *L. leucocephala* (60:20:20); R4, *P. purpureum* + concentrate + *G. sepium* + *L. leucocephala* (60:20:10:10)

CP, crude protein; DM, dry matter; RUP, ruminally undegraded protein; RDP, ruminally degraded protein; Sol P, soluble protein; ME, metabolizable energy; NEI, net energy lactation; NDF, neutral detergent fiber; peNDF, physically effective neutral detergent fiber; NFC, non fiber carbohydrate; Sol fiber, soluble fiber; EE, ether extract; LCFA, long chain fatty acids; Ca, calcium; P, phosphorus

Table 3. Rumen Values of The Simulated Treatments at Different Environmental Temperatures

| Component | R1 | | R2 | | R3 | | R4 | |
|------------------------|-------|-------|-------|-------|-------|-------|-------|-------|
| | 20 °C | 30 °C | 20 °C | 30 °C | 20 °C | 30 °C | 20 °C | 30 °C |
| Total FC bacteria (g) | 871 | 871 | 606 | 606 | 660 | 660 | 633 | 633 |
| Total NFC bacteria (g) | 399 | 399 | 1134 | 1134 | 704 | 704 | 921 | 921 |
| Total bacteria (g) | 1270 | 1270 | 1740 | 1740 | 1364 | 1364 | 1554 | 1554 |
| Bacterial N (g) | 127 | 127 | 174 | 174 | 136 | 136 | 155 | 155 |
| RUP N (g) | 104 | 104 | 141 | 141 | 141 | 141 | 140 | 140 |
| Rumen pH | 6.46 | 6.46 | 6.46 | 6.46 | 6.46 | 6.46 | 6.46 | 6.46 |

R1, *P. purpureum*; R2, *P. purpureum* + concentrate (60:40); R3, *P. purpureum* + *G. sepium* + *L. leucocephala* (60:20:20); R4, *P. purpureum* + concentrate + *G. sepium* + *L. leucocephala* (60:20:10:10)

FC, fiber carbohydrate; NFC, non fiber carbohydrate; RUP, ruminally undegraded protein

ferment only cell wall carbohydrate, use ammonia only as a N source, and do not ferment peptides or amino acids. On the other hand, the NFC bacteria ferment non structural carbohydrates (starch, pectin, sugars, etc.), use either ammonia or peptides and amino acids as an N source, and can produce ammonia (Russell *et al.*, 1992).

Total FC bacteria were highest in R1 (871 g) compared to other treatments (varied between 606-660 g) since it contained the highest NDF. However, total NFC bacteria of R1 were lowest. The highest NFC bacteria was found after supplementing *P. purpureum* (R1) with concentrate in the amount of 40% DM. The concentrate supplementation increased total NFC bacteria from 399 to 1134 g. Similar response was found when concentrate replaced 50% of the leguminous leaves supplementation treatment (R4 compared to R3). This was due to high proportion of sugar, starch, crude protein (CP) and ruminally degraded protein (RDP) in the concentrate mixture. Under supplementary feeding condition, it was clear that total bacteria mass or bacterial N was determined more by the amount of NFC bacteria rather than that of FC bacteria. It has been well-known that microbes fermenting starch, pectin and sugars (NFC) grow more rapidly than those which fermenting structural carbohydrate or FC microbes, provided that a suitable N source is available (Russell and Hespell, 1981; Fox *et al.*, 2004).

Treatments R2, R3 and R4 had similar amount of RUP N (ruminally undegraded protein nitrogen), i.e. 140-141 g. These might be explained by the similarities of crude protein contents and RUP proportions between R2, R3 and R4. These figures were contrast with R1 where the RUP N was

substantially lower, i.e. 104 g. The RUP N value for R1 may low since the absolute amount of protein entering the rumen is already small (no supplementation). Protein can escape from the rumen since peptides arising from protein degradation are only utilized by ruminal microorganisms at a limited rate, i.e. 0.07 g of peptide per gram of microorganism per hour. When the degradation of the protein is rapid, peptides accumulate and a portion of peptides as well as protein may escape (Sniffen *et al.*, 1992). It is interesting to note that the rumen sub-model of the CNCPS does not have a protozoal pool although protozoa can comprise as much as 50% of the microbial mass in the rumen. This might be due to the work of Weller and Pilgrim (1974) who noted that protozoa lyse easily, recycle themselves, and contribute little microbial protein to the animal.

All simulated treatments have similar pH value according to the CNCPS model, i.e. 6.46. The CNCPS predicts rumen pH from physical characteristics of feeds as related to their effectiveness in stimulating chewing, rumination and increased rumen motility based on their total cell wall content and particle size within classes of feeds or by physically effective NDF (peNDF) contents (Tylutki *et al.*, 2008). The equation only distinguish between peNDF less than 24.5% DM and above it. Above this threshold peNDF value, rumen pH is assumed to have pH 6.46 (Fox *et al.*, 2004). Since the peNDF of all simulated treatments were higher than 24.5%, hence, the pH values of all treatments were the same. The peNDF values of R1, R2, R3 and R4 were 44.40, 30.40, 45.30 and 37.85% DM, respectively. Fox *et al.* (2004) discussed limitations of the CNCPS approach to predict rumen pH. The

CNCPS does not attempt to integrate ruminal pH with the rate or amount of NFC digestion, and effects of ruminal fluid dilution rate on VFA removal from the rumen are not considered. It might be worthy to develop a more integrated rumen model, in which microbial growth is more integrated with digestion and passage and rumen concentrations of VFA are used to predict pH.

There was no difference between different environmental temperatures on all rumen value variables at each simulated treatment. The CNCPS does not include the variation of environmental temperature into microbial growth and rumen metabolism models. This may address a limitation of the model since environmental temperature significantly affects rumen function and metabolism as reported by some authors (Gengler *et al.*, 1970; Bernabucci *et al.*, 1999; Arieli *et al.*, 2004). Different environmental temperatures significantly changed acetic, propionic and butyric acids production, and rumen temperature (Gengler *et al.*, 1970), affected fiber (NDF and ADF) digestibility, rumen passage rate (Bernabucci *et al.*, 1999), rumen ammonia and milk urea nitrogen (Arieli *et al.*, 2004). Therefore, improvement of the current model by integrating ambient temperature variation to obtain more accurate result is needed.

Nutrient Balance

Nutrient balance refers to the difference between nutrient supply from the feeds and nutrient requirement of the animal. Nutrient supply is originated from the quantity (amount) and quality (availability) of each nutrient in the feed consumed by the animal. Nutrient requirement of a dairy cow can be divided into maintenance, growth, pregnancy and lactation requirements (Fox *et al.*, 1992). In the present paper, the nutrient balance variables are presented in the form of percent required rather than the absolute balance values. This may give additional information on how far the nutrient supply meets the animal requirement in a specific production level.

The nutrient balance of the simulated treatments at different environmental temperatures is presented in Table 4. Based on the CNCPS model, there were negative balances of metabolizable energy (ME) and metabolizable protein (MP) if a lactating dairy cow fed only by *P. purpureum* (R1) at 20 °C and 30 °C. The ME balance was worse at higher temperature (decreased from 55% to 49% of the requirement), while the MP balance was remain unchanged (71% of the requirement). Concentrate supplementation (R2) provided adequate ME and MP balances at 20 °C, but the ME balance was slightly below the requirement at 30 °C. Higher ME, CP and soluble protein contents in the ration might be the reason of

Table 4. Nutrient Balance of The Simulated Treatments at Different Environmental Temperatures

| Component | R1 | | R2 | | R3 | | R4 | |
|-------------------------------|-------|-------|-------|-------|-------|-------|-------|-------|
| | 20 °C | 30 °C | 20 °C | 30 °C | 20 °C | 30 °C | 20 °C | 30 °C |
| ME (% Req) | 55 | 49 | 101 | 91 | 68 | 61 | 85 | 77 |
| MP (% Req) | 71 | 71 | 106 | 106 | 80 | 80 | 92 | 92 |
| NH ₃ -N (% Req) | 116 | 116 | 150 | 150 | 174 | 174 | 162 | 162 |
| Peptide N (% Req) | 197 | 197 | 139 | 139 | 220 | 220 | 170 | 170 |
| Lysine (% Req) | 75 | 75 | 129 | 129 | 91 | 91 | 109 | 109 |
| Methionine (% Req) | 93 | 93 | 155 | 155 | 102 | 102 | 128 | 128 |
| peNDF (% Req) | 193 | 193 | 132 | 132 | 197 | 197 | 165 | 165 |
| Ca (% Req) | 69 | 69 | 133 | 133 | 103 | 103 | 118 | 118 |
| P (% Req) | 93 | 93 | 205 | 205 | 69 | 69 | 137 | 137 |
| Total ME available (Mcal/day) | 21.1 | 21.1 | 31.7 | 31.7 | 23.7 | 23.7 | 27.7 | 27.7 |

R1, *P. purpureum*; R2, *P. purpureum* + concentrate (60:40); R3, *P. purpureum* + *G. sepium* + *L. leucocephala* (60:20:20); R4, *P. purpureum* + concentrate + *G. sepium* + *L. leucocephala* (60:20:10:10) ME, metabolizable energy; MP, metabolizable protein; peNDF, physically effective neutral detergent fiber; Ca, calcium; P, phosphorus %Req, percent required

such response. This was confirmed that concentrate supplementation increased nutrient flow and milk production of dairy cows (Bargo *et al.*, 2002; Sairanen *et al.*, 2005; McEvoy *et al.*, 2008). Supplementation of leguminous tree leaves (R3) did not improve much total ME available compared to R1 (23.7 vs 21.1 Mcal/day) and, hence, negative energy balance occurred. The same pattern was observed for the MP balance. These were due to lower nutrient supply from the respective leguminous tree leaves compared to the concentrate. The results were opposed with the results from experiments conducted by Richards *et al.* (1994) and Ondiek *et al.* (2000) using lactating dairy goats. They concluded that *G. sepium* and *L. leucocephala* could contribute as nitrogen sources in compounded diet supplements without any detrimental effects on production in dairy goats (Ondiek *et al.*, 2000), and up to 50% of the concentrate N may be replaced by the tropical tree legumes gliricidia and leucaena without a reduction in milk production (Richards *et al.*, 1994). Combination of all supplements, i.e. concentrate and legume leaves (R4) performed better ME and MP balances, but remained not better than the concentrate supplementation only (R2).

Hot weather has been known to affect animal bioenergetics, with adverse effects on the performance and well being of livestock. Reduced feed intake, growth or milk production, efficiency and reproduction are recognized results (Thatcher, 1974; Hahn, 1999). Hahn (1999) also showed that the threshold temperature for thermoneutral (comfortable) and hot stress was approximately 25 °C. The CNCPS itself stated that the temperature of 20 °C was considered to have no effect on basal metabolic rate and there is no cold or heat stress at that temperature. Thus, the temperature of 20 °C was described as being thermoneutral (Fox and Tylutki, 1998). This was the reason why we chose the temperatures at 20 °C and 30 °C, i.e. to compare nutrient balance under comfortable condition and under hot stress condition at different supplementary feeding strategies.

In the CNCPS environment, it is assumed that the effect of environmental temperature is primarily reflected through a change in the requirement for maintenance energy and dry matter intake (DMI) and that the impact on requirements for pregnancy, growth

and lactation are secondary to energy available after the maintenance requirement is met (Fox and Tylutki, 1998). Therefore, based on this assumption, environmental temperature was associated only with energy balance in the present research since DMI was inputted at a constant value of 15 kg/day, and no association with the MP balance. Maintenance energy requirement is higher if the ambient temperature falls outside the thermoneutral temperature, i.e. both under cold stress and heat stress (Fox and Tylutki, 1998). Hence, it was not surprising that the ME balance became lower at 30 °C compared to 20 °C at all feed treatments.

All simulated treatments including the non supplemented one (R1) had positive NH₃-N and peptide N balances in the rumen. This suggested sufficient N supply for optimal microbial growth. Ammonia N and peptide N balances were the highest for R3, i.e. 174 and 220% of the requirements, respectively, although the CP and RDP contents were lower than R2. This was due to the influence of rate of passage. Low fiber feeds such as concentrate have relatively high rate of passage (Sairanen *et al.*, 2005) and this is correlated to its low peNDF value (Fox *et al.*, 2004). The fact implies that the protein from concentrate is more available post-ruminally while the protein from leguminous leaves is more available in the rumen. Similar explanation is applied of the reduced values of NH₃-N and peptide N balances when concentrate supplement substituted 50% of the legume supplement (R4 compared to R3).

Concentrate supplementation (R2 and R4) fulfilled lysine and methionine requirements. However, supplementation of *G. sepium* and *L. leucocephala* without concentrate (R3) did not meet the requirement. Protein from plant origin generally contains low lysine content, as opposed to protein from animal origin which is high in lysine (McDonald *et al.*, 2002). Since the concentrate contained a portion of fish meal as a protein source, therefore, it contributed to the lysine requirement of lactating dairy cow model. Concentrate used in the present research also contained dicalcium phosphate (DCP) as calcium (Ca) and phosphorus (P) sources, and led to adequate Ca and P supply for treatments with concentrate supplementation (R2 and R4). On the other hand, leguminous leaves supplementation (R3) led to insufficient P supply due to its low content in the

leaves especially in *G. sepium*. It seems necessary, therefore, to add P source when supplementation is based on leguminous leaves.

Manure Excretion

Manure excretion data of the simulated treatments at different ambient temperatures are presented in Table 5. Manure refers to fecal and urine excreta. There was almost no difference in total manure production for all simulated treatments (centered at approximately 50 kg of manure). Total manure N was highest in R2, followed by R4, R3 and R1. This suggested that the more N supply from the ration, the more N is excreted. Nennich *et al.* (2005) showed a linear relationship between crude protein intake and

animal is trying to use and metabolize available N more efficiently.

Similar to N pattern, the more P supply from feeds led to the more total manure P. The R3 treatment had the highest percentage of productive P (41%), followed by R1, R4 and R2 with the percentage of 31, 22 and 15%, respectively. Since R3 had negative P balance, hence, animal used the available P in more efficient manner. Therefore, it is important not to give N and P in excessive amount to maintain high partitioning of the respective elements for productive purpose. Moreover, reduced level of N and P in the manure is more environmentally friendly and less contamination to the environment (Lanyon, 1994; Newton *et al.*, 2003). Precision ration

Table 5. Manure Excretion of The Simulated Treatments at Different Environmental Temperatures

| Component | R1 | | R2 | | R3 | | R4 | |
|--------------------------|-------|-------|-------|-------|-------|-------|-------|-------|
| | 20 °C | 30 °C | 20 °C | 30 °C | 20 °C | 30 °C | 20 °C | 30 °C |
| Total manure (kg) | 50 | 50 | 47 | 47 | 52 | 52 | 49 | 49 |
| Total manure N (g) | 86 | 86 | 224 | 224 | 195 | 195 | 210 | 210 |
| Productive N/total N (%) | 53 | 53 | 28 | 28 | 31 | 31 | 30 | 30 |
| Manure N/total N (%) | 47 | 47 | 72 | 72 | 69 | 69 | 70 | 70 |
| Total manure P (g) | 29 | 29 | 72 | 72 | 18 | 18 | 45 | 45 |
| Productive P/total P (%) | 31 | 31 | 15 | 15 | 41 | 41 | 22 | 22 |
| Manure P/total P (%) | 69 | 69 | 85 | 85 | 59 | 59 | 78 | 78 |

R1, *P. purpureum*; R2, *P. purpureum* + concentrate (60:40); R3, *P. purpureum* + *G. sepium* + *L. leucocephala* (60:20:20); R4, *P. purpureum* + concentrate + *G. sepium* + *L. leucocephala* (60:20:10:10) N, nitrogen; P, phosphorus

N excretion, which support the argument. Chen *et al.* (2008) also used N feed content to predict manure N content using artificial neural network approach, beside the psychochemical properties of the manure, and the findings demonstrated that the predictors (N feed content and psychochemical properties) and the model were appropriate to predict dairy manure N content. Regarding the partitioning of productive N or manure N to total N, treatment of *P. purpureum* without supplementation (R1) was the most productive with value of 53%. Supplementation treatments (R2 to R4) had productive N to total N percentage around 30%, and the rest went to manure. It seemed that when there is N supply limitation,

formulation and feeding should be encouraged for benefiting from such aspect.

CONCLUSION

Supplementary feeding is necessary for improving the nutrient balance of lactating dairy cow, especially when the cow is maintained under uncomfortable environmental temperature. Both concentrate and leguminous tree leaves (*G. sepium* and *L. leucocephala*) can be used as supplements to fulfill nutrient balance or at least to minimize negative nutrient balance. Under higher environmental temperature, maintenance energy requirement was higher and contributed to more severe negative

energy balance. Supplementation increased total bacteria and bacterial N supplies compared to the control treatment. On the other hand, supplementation increased the proportion and absolute amount of N and P excreted as a consequence of increased nutrient supply. Comparing the concentrate and the legume supplements, concentrate was superior compared to the legume leaves to provide adequate nutrient supply. However, the cost associated with buying of concentrate should also be taken into account. Mixture of commercial concentrate with locally available leguminous leaves may give an optimum ration and maximizing benefit in term of balancing between production and costs.

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